

=====

Sequence Listing was accepted.

If you need help call the Patent Electronic Business Center at (866)  
217-9197 (toll free).

Reviewer: Durreshwar Anjum

Timestamp: [year=2010; month=3; day=30; hr=14; min=53; sec=12; ms=838; ]

=====

Application No: 10587723

Version No: 1.0

Input Set:

Output Set:

Started: 2010-03-23 14:30:29.458

Finished: 2010-03-23 14:30:33.533

Elapsed: 0 hr(s) 0 min(s) 4 sec(s) 75 ms

Total Warnings: 212

Total Errors: 0

No. of SeqIDs Defined: 212

Actual SeqID Count: 212

Error code	Error Description
W 213	Artificial or Unknown found in <213> in SEQ ID (1)
W 213	Artificial or Unknown found in <213> in SEQ ID (2)
W 213	Artificial or Unknown found in <213> in SEQ ID (3)
W 213	Artificial or Unknown found in <213> in SEQ ID (4)
W 213	Artificial or Unknown found in <213> in SEQ ID (5)
W 213	Artificial or Unknown found in <213> in SEQ ID (6)
W 213	Artificial or Unknown found in <213> in SEQ ID (7)
W 213	Artificial or Unknown found in <213> in SEQ ID (8)
W 213	Artificial or Unknown found in <213> in SEQ ID (9)
W 213	Artificial or Unknown found in <213> in SEQ ID (10)
W 213	Artificial or Unknown found in <213> in SEQ ID (11)
W 213	Artificial or Unknown found in <213> in SEQ ID (12)
W 213	Artificial or Unknown found in <213> in SEQ ID (13)
W 213	Artificial or Unknown found in <213> in SEQ ID (14)
W 213	Artificial or Unknown found in <213> in SEQ ID (15)
W 213	Artificial or Unknown found in <213> in SEQ ID (16)
W 213	Artificial or Unknown found in <213> in SEQ ID (17)
W 213	Artificial or Unknown found in <213> in SEQ ID (18)
W 213	Artificial or Unknown found in <213> in SEQ ID (19)
W 213	Artificial or Unknown found in <213> in SEQ ID (20)

**Input Set:**

**Output Set:**

**Started:** 2010-03-23 14:30:29.458  
**Finished:** 2010-03-23 14:30:33.533  
**Elapsed:** 0 hr(s) 0 min(s) 4 sec(s) 75 ms  
**Total Warnings:** 212  
**Total Errors:** 0  
**No. of SeqIDs Defined:** 212  
**Actual SeqID Count:** 212

Error code

Error Description

This error has occurred more than 20 times, will not be displayed

# SEQUENCE LISTING

<110> MILLER, JEFFREY C.  
ZHANG, LEI

<120> METHODS AND COMPOSITIONS FOR TARGETED CLEAVAGE AND RECOMBINATION

<130> 8325-0036.31

<140> 10587723

<141> 2010-03-23

<150> PCT/US04/25407

<151> 2004-08-06

<150> 60/542,780

<151> 2004-02-05

<150> 60/556,831

<151> 2004-03-26

<150> 60/575,919

<151> 2004-06-01

<150> 10/912,932

<151> 2004-08-06

<160> 212

<170> PatentIn version 3.3

<210> 1

<211> 44

<212> DNA

<213> Artificial

<220>

<223> human hSMC1L1 gene

<220>

<221> misc\_feature

<222> (5)..(16)

<223> Target sequence for the hSMC1-specific ZFP

<220>

<221> misc\_feature

<222> (23)..(34)

<223> Target sequence for the hSMC1-specific ZFP

<400> 1

ctgccgcgcg cgccgcggcc gtcattgggt tcttgaaact gatt

44

<210> 2

<211> 7

<212> PRT  
<213> Artificial

<220>  
<223> human hSMC1L1 gene

<400> 2

Met Gly Phe Leu Lys Leu Ile  
1 5

<210> 3  
<211> 47  
<212> DNA  
<213> Artificial

<220>  
<223> human x chromosome SMC1 region

<400> 3  
ctgccgccgg cgccgcggcc gtcattggggt tcctgaaact gattgag

47

<210> 4  
<211> 8  
<212> PRT  
<213> Artificial

<220>  
<223> human x chromosome SMC1 region

<400> 4

Met Gly Phe Leu Lys Leu Ile Glu  
1 5

<210> 5  
<211> 50  
<212> DNA  
<213> Artificial

<220>  
<223> artificial donor oligonucleotide

<400> 5  
ctgccgccgg cgccgcggcc gtcataagaa gcttcctgaa actgattgag

50

<210> 6  
<211> 463  
<212> DNA  
<213> Artificial

<220>  
<223> an amplification product derived from a mutated hSMC1 gene

<400> 6  
tagtcctgca ggttttaaagc aattcgccct tctcagcaag cgtgagctca ggtctccccc 60  
gcctccttga acctcaagaa ctgctctgac tccgcccagc aacaactcct ccggggatct 120  
ggtccgcagg agcaagtgtt tgttggtgcc atgcaacaag aaaagggggc ggaggcacca 180  
cgccagtcgt cagctcgctc ctcgtatacg caacatcagt ccccgccctt ggtccactc 240  
ctgccggaag gcgaagatcc cgtaggcct ggacgtatc tcgcgacatt tgccggctgc 300  
ccggcttgca ctgcggcggt tcccgcgcg gctacctcag ttctcgggcg tacggcgcg 360  
cctgtcctac tgctgccggc gcccgggccg tcataagaag cttcctgaaa ctgattgaag 420  
ggcgaattcg cggcgctaa attcaattcg ccctatagtg agt 463

<210> 7  
<211> 50  
<212> DNA  
<213> Artificial

<220>  
<223> human IL2Rgamma gene

<220>  
<221> misc\_feature  
<222> (8)..(16)  
<223> Target sequence for the second pair of IL2Rgamma-specific ZFP

<220>  
<221> misc\_feature  
<222> (23)..(34)  
<223> Target sequence for the second pair of IL2Rgamma-specific ZFP

<400> 7  
cttccaacct ttctcctcta ggtacaagaa ctcggataat gataaagtcc 50

<210> 8  
<211> 9  
<212> PRT  
<213> Artificial

<220>  
<223> human IL2Rgamma gene

<400> 8

Tyr Lys Asn Ser Asp Asn Asp Lys Val  
1 5

<210> 9

<211> 59  
 <212> DNA  
 <213> Artificial  
  
 <220>  
 <223> IL2Rgamma gene  
  
 <400> 9  
 gttcctcttc cttccaacct ttctcctcta ggtacaagaa ctcgataat gataaagtc 59  
  
 <210> 10  
 <211> 9  
 <212> PRT  
 <213> Artificial  
  
 <220>  
 <223> IL2Rgamma gene  
  
 <400> 10  
  
 Tyr Lys Asn Ser Asp Asn Asp Lys Val  
 1 5  
  
 <210> 11  
 <211> 59  
 <212> DNA  
 <213> Artificial  
  
 <220>  
 <223> artificial donor oligonucleotide  
  
 <400> 11  
 gttcctcttc cttccaacct ttctcctcta ggtaaaagaa ttccgacaac gataaagtc 59  
  
 <210> 12  
 <211> 624  
 <212> DNA  
 <213> Artificial  
  
 <220>  
 <223> mutated IL2Rgamma gene  
  
 <400> 12  
 tagtcctgca ggtttaaacg aattcgccct ttctctagg taaaagaatt ccgacaacga 60  
  
 taaagtccag aagtgcagcc actatctatt ccctgaagaa atcacttctg gctgtcagtt 120  
  
 gcaaaaaaag gagatccacc tctaccaaac atttgttggt cagctccagg acccacggga 180  
  
 acccaggaga caggccacac agatgctaaa actgcagaat ctgggtaatt tggaaagaaa 240  
  
 ggggtcaagag accagggata ctgtgggaca ttggagtcta cagagtagtg ttcttttatc 300  
  
 ataagggtac atgggcagaa aagaggaggt aggggatcat gatgggaagg gaggaggtat 360

taggggcact accttcagga tcctgacttg tctaggccag gggaatgacc acatatgcac	420
acatatctcc agtgatcccc tgggctccag agaacctaac acttcacaaa ctgagtgaat	480
cccagctaga actgaactgg aacaacagat tcttgaacca ctgtttggag cacttggtgc	540
agtaccggac taagggcgaa ttgcggccg ctaaattcaa ttgcacctat agtgagtcgt	600
attacaattc actggccgtc gttt	624

<210> 13  
 <211> 700  
 <212> DNA  
 <213> Artificial

<220>  
 <223> human beta-globin gene

<400> 13	
tactgatggt atggggccaa gagatatatc ttagagggag ggctgagggt ttgaagtcca	60
actcctaagc cagtgccaga agagccaagg acaggtacgg ctgtcatcac ttagacctca	120
ccctgtggag ccacacctta gggttggcca atctactccc aggagcaggg agggcaggag	180
ccagggctgg gcataaaagt cagggcagag ccactctattg cttacatttg cttctgacac	240
aactgtgttc actagcaacc tcaaacagac accatggtgc atctgactcc tgaggagaag	300
tctgccgtta ctgcctctg gggcaagggtg aacgtggatg aagttggtgg tgaggccctg	360
ggcaggttgg tatcaagggtt acaagacagg ttttaaggaga ccaatagaaa ctgggcatgt	420
ggagacagag aagactcttg ggtttctgat aggcactgac tctctctgcc tatttgtcta	480
ttttccacc cttaggctgc tgggtgtcta cccttgacc cagaggttct ttgagtcctt	540
tggggatctg tccactcctg atgctgttat gggcaaccct aaggtgaagg ctcatggcaa	600
gaaagtgtc ggtgccttta gtgatggcct ggctcacctg gacaacctca agggcacctt	660
tgccacactg agtgagctgc actgtgacaa gctgcacgtg	700

<210> 14  
 <211> 408  
 <212> DNA  
 <213> Artificial

<220>  
 <223> an amplification product derived from a mutated beta-globin gene

<400> 14	
tgcttaccaa gctgtgattc caaatattac gtaaatacac ttgcaaagga ggatgttttt	60



agtagcaatt tgtactgatg gtatggggcc aagagatata tcttagaggg agggctgagg 120  
 gtttgaagtc caactcctaa gccagtgcc aagagagccaa ggacaggtag ggctgtcatc 180  
 acttagacct caccctgtgg agccacaccc tagggttggc caatctactc ccaggagcag 240  
 ggagggcagg agccagggct gggcataaaa gtcagggcag agccatctat tgcttacatt 300  
 tgcttctgac acaactgtgt tcactagcaa cctcaaacag acaccatggt gcctctgact 360  
 cctgaggaga agtctggcgt tagtgcccga attccgatcg tcaaccac 408

<210> 15  
 <211> 42  
 <212> DNA  
 <213> Artificial

<220>  
 <223> IL-2Rgamma gene

<220>  
 <221> misc\_feature  
 <222> (1)..(13)  
 <223> target sequences for the 5-10 ZFP/FokI fusion protein

<220>  
 <221> misc\_feature  
 <222> (31)..(42)  
 <223> target sequences for the 5-8 ZFP/FokI fusion protein

<400> 15  
 cacgtttcgt gttcggagcc gctttaaccc actctgtgga ag 42

<210> 16  
 <211> 336  
 <212> PRT  
 <213> Artificial

<220>  
 <223> 5-8 ZFP/FokI fusion

<220>  
 <221> MISC\_FEATURE  
 <222> (1)..(17)  
 <223> NLS

<220>  
 <221> MISC\_FEATURE  
 <222> (18)..(130)  
 <223> ZFP

<220>  
 <221> MISC\_FEATURE

<222> (131)..(140)

<223> ZC linker

<220>

<221> MISC\_FEATURE

<222> (141)..(336)

<223> FokI cleavage half-domain

<400> 16

Met Ala Pro Lys Lys Lys Arg Lys Val Gly Ile His Gly Val Pro Ala  
1 5 10 15

Ala Met Ala Glu Arg Pro Phe Gln Cys Arg Ile Cys Met Arg Asn Phe  
20 25 30

Ser Arg Ser Asp Asn Leu Ser Glu His Ile Arg Thr His Thr Gly Glu  
35 40 45

Lys Pro Phe Ala Cys Asp Ile Cys Gly Arg Lys Phe Ala Arg Asn Ala  
50 55 60

His Arg Ile Asn His Thr Lys Ile His Thr Gly Ser Gln Lys Pro Phe  
65 70 75 80

Gln Cys Arg Ile Cys Met Arg Asn Phe Ser Arg Ser Asp Thr Leu Ser  
85 90 95

Glu His Ile Arg Thr His Thr Gly Glu Lys Pro Phe Ala Cys Asp Ile  
100 105 110

Cys Gly Arg Lys Phe Ala Ala Arg Ser Thr Arg Thr Thr His Thr Lys  
115 120 125

Ile His Leu Arg Gln Lys Asp Ala Ala Arg Gly Ser Gln Leu Val Lys  
130 135 140

Ser Glu Leu Glu Glu Lys Lys Ser Glu Leu Arg His Lys Leu Lys Tyr  
145 150 155 160

Val Pro His Glu Tyr Ile Glu Leu Ile Glu Ile Ala Arg Asn Ser Thr  
165 170 175

Gln Asp Arg Ile Leu Glu Met Lys Val Met Glu Phe Phe Met Lys Val  
180 185 190

Tyr Gly Tyr Arg Gly Lys His Leu Gly Gly Ser Arg Lys Pro Asp Gly  
195 200 205

Ala Ile Tyr Thr Val Gly Ser Pro Ile Asp Tyr Gly Val Ile Val Asp  
210 215 220

Thr Lys Ala Tyr Ser Gly Gly Tyr Asn Leu Pro Ile Gly Gln Ala Asp  
225 230 235 240

Glu Met Gln Arg Tyr Val Glu Glu Asn Gln Thr Arg Asn Lys His Ile  
245 250 255

Asn Pro Asn Glu Trp Trp Lys Val Tyr Pro Ser Ser Val Thr Glu Phe  
260 265 270

Lys Phe Leu Phe Val Ser Gly His Phe Lys Gly Asn Tyr Lys Ala Gln  
275 280 285

Leu Thr Arg Leu Asn His Ile Thr Asn Cys Asn Gly Ala Val Leu Ser  
290 295 300

Val Glu Glu Leu Leu Ile Gly Gly Glu Met Ile Lys Ala Gly Thr Leu  
305 310 315 320

Thr Leu Glu Glu Val Arg Arg Lys Phe Asn Asn Gly Glu Ile Asn Phe  
325 330 335

<210> 17  
<211> 339  
<212> PRT  
<213> Artificial

<220>  
<223> 5-10 ZFP/FokI fusion

<220>  
<221> MISC\_FEATURE  
<222> (1)..(17)  
<223> NLS

<220>  
<221> MISC\_FEATURE  
<222> (18)..(133)  
<223> ZFP

<220>

<221> MISC\_FEATURE

<222> (134)..(143)

<223> ZC linker

<220>

<221> MISC\_FEATURE

<222> (144)..(339)

<223> FokI cleavage half-domain

<400> 17

Met Ala Pro Lys Lys Lys Arg Lys Val Gly Ile His Gly Val Pro Ala  
1 5 10 15

Ala Met Ala Glu Arg Pro Phe Gln Cys Arg Ile Cys Met Arg Asn Phe  
20 25 30

Ser Arg Ser Asp Ser Leu Ser Arg His Ile Arg Thr His Thr Gly Glu  
35 40 45

Lys Pro Phe Ala Cys Asp Ile Cys Gly Arg Lys Phe Ala Asp Ser Ser  
50 55 60

Asn Arg Lys Thr His Thr Lys Ile His Thr Gly Gly Gly Gly Ser Gln  
65 70 75 80

Lys Pro Phe Gln Cys Arg Ile Cys Met Arg Asn Phe Ser Arg Ser Asp  
85 90 95

Ser Leu Ser Val His Ile Arg Thr His Thr Gly Glu Lys Pro Phe Ala  
100 105 110

Cys Asp Ile Cys Gly Arg Lys Phe Ala Asp Arg Ser Asn Arg Ile Thr  
115 120 125

His Thr Lys Ile His Leu Arg Gln Lys Asp Ala Ala Arg Gly Ser Gln  
130 135 140

Leu Val Lys Ser Glu Leu Glu Glu Lys Lys Ser Glu Leu Arg His Lys  
145 150 155 160

Leu Lys Tyr Val Pro His Glu Tyr Ile Glu Leu Ile Glu Ile Ala Arg  
165 170 175

Asn Ser Thr Gln Asp Arg Ile Leu Glu Met Lys Val Met Glu Phe Phe  
180 185 190

Met Lys Val Tyr Gly Tyr Arg Gly Lys His Leu Gly Gly Ser Arg Lys  
 195 200 205

Pro Asp Gly Ala Ile Tyr Thr Val Gly Ser Pro Ile Asp Tyr Gly Val  
 210 215 220

Ile Val Asp Thr Lys Ala Tyr Ser Gly Gly Tyr Asn Leu Pro Ile Gly  
 225 230 235 240

Gln Ala Asp Glu Met Gln Arg Tyr Val Glu Glu Asn Gln Thr Arg Asn  
 245 250 255

Lys His Ile Asn Pro Asn Glu Trp Trp Lys Val Tyr Pro Ser Ser Val  
 260 265 270

Thr Glu Phe Lys Phe Leu Phe Val Ser Gly His Phe Lys Gly Asn Tyr  
 275 280 285

Lys Ala Gln Leu Thr Arg Leu Asn His Ile Thr Asn Cys Asn Gly Ala  
 290 295 300

Val Leu Ser Val Glu Glu Leu Leu Ile Gly Gly Glu Met Ile Lys Ala  
 305 310 315 320

Gly Thr Leu Thr Leu Glu Glu Val Arg Arg Lys Phe Asn Asn Gly Glu  
 325 330 335

Ile Asn Phe

<210> 18

<211> 797

<212> DNA

<213> Artificial

<220>

<223> enhanced Green Fluorescent Protein gene

<400> 18

cgaattctgc agtcgacggt accgcggggcc cgggatccac cggtcgccac catggtgagc 60

aagggcgagg agctgttcac cgggggtggtg cccatcctgg tcgagctgga cggcgacgta 120

aacggccaca agttcagcgt gtccggcgag ggcgagggcg atgccaccta cggcaagctg 180

accctgaagt tcatctgcac caccggcaag ctgcccgtagc cctggcccac cctcgtgacc	240
accctgacct acggcggtgca gtgcttcagc cgctaccccg accacatgaa gcagcacgac	300
ttcttcaagt cggccatgcc cgaaggctac gtccaggagc gcaccatctt cttcaaggac	360
gacggcaact acaagacccg cgccgaggtg aagttcgagg gcgacaccct ggtgaaccgc	420
atcgagctga agggcatcga cttcaaggag gacggcaaca tcctggggca caagctggag	480
tacaactaca acagccacaa cgtctatatc atggccgaca agcagaagaa cggcatcaag	540
gtgaacttca agatccgcca caacatcgag gacggcagcg tgcagctcgc cgaccactac	600
cagcagaaca ccccatcgg cgacggcccc gtgctgctgc ccgacaacca ctacctgagc	660
accagtcgg cctgagcaa agaccccaac gagaagcgcg atcacatggc cctgctggag	720
ttcgtgaccg cggccgggat cactctcggc atggacgagc tgtacaagta aagcggccgc	780
gactctagat cataatc	797

<210> 19

<211> 795

<212> DNA

<213> Artificial

<220>

<223> mutant defective eGFP gene

<400> 19

cgaattctgc agtcgacggc accgcggggc cgggataccac cggtcgccac catggtgagc	60
aaggcgagag agctgttcac cgggggtggtg cccatcctgg tcgagctgga cggcgacgta	120
aacggccaca agttcagcgt gtccggcgag ggcgagggcg atgccacctt cggcaagctg	180
accctgaagt tcatctgcac caccggcaag ctgcccgtagc cctggcccac cctcgtgacc	240
accctgacct acggcggtgca gtgcttcagc cgctacccct aacacgaagc agcacgactt	300
cttcaagtcc gccatgcccg aaggctacgt ccaggagcgc accatcttct tcaaggacga	360
cggcaactac aagaccgcg ccgaggtgaa gttcgagggc gacaccctgg tgaaccgcat	420
cgagctgaag ggcacgact tcaaggagga cggcaacatc ctggggcaca agctggagta	480
caactacaac agccacaacg tctatatcat ggccgacaag cagaagaacg gcatcaaggt	540
gaacttcaag atccgccaca acatcgagga cggcagcgtg cagctcgccg accactacca	600
gcagaacacc cccatcgggc acggccccgt gctgctgccc gacaaccact acctgagcac	660
ccagtccgcc ctgagcaaag accccaacga gaagcgcgat cacatggtcc tgctggagtt	720
cgtgaccgcc gccgggatca ctctcggcac ggacgagctg tacaagtaa gcggccgcga	780

ctctagatca taatc

795

<210> 20

<211> 734

<212> DNA

<213> Artificial

<220>

<223> eGFP insert in pCR(R)4-TOPO-GFPdonor5

<400> 20

ggcgaggagc tgttcaccgg ggtggtgccc atcctggtcg agctggacgg cgacgtaaac 60

ggccacaagt tcagcgtgtc cggcgagggc gagggcgatg ccacctacgg caagctgacc 120

ctgaagttca tctgcaccac cggcaagctg cccgtgccct ggcccaccct cgtgaccacc 180

ctgacctacg gcggtgcagtg cttcagccgc taccctgacc acatgaagca gcacgacttc 240

ttcaagtccg ccatgcccga aggctacgtc caggagcgca ccatcttctt caaggacgac 300

ggcaactaca agaccgcgc cgaggtgaag ttcgagggcg acaccctggt gaaccgcatc 360

gagctgaagg gcatcgactt caaggaggac ggcaacatcc tggggcacia gctggagtac 420

aactacaaca gccacaacgt ctatatcatg gccgacaagc agaagaacgg catcaagggtg 480

aacttcaaga tccgccacia catcgaggac ggacgcgtgc agctcgccga ccactaccag 540

cagaacaccc ccatcggcga cgccccgtg ctgctgcccg acaaccacta cctgagcacc 600

cagtcgccc tgagcaaaga ccccaacgag aagcgcgac acatggtcct gctggagttc 660

gtgaccgccc cgggatcac tctcgcatg gacgagctgt acaagtaaag cggccgcgac 720

tctagatcat aatc 734

<210> 21

<211> 1527

<212> DNA

<213> Artificial

<220>

<223> eGFP insert in pCR(R)4-TOPO

<400> 21

ggcgaggagc tgttcaccgg ggtggtgccc atcctggtcg agctggacgg cgacgtaaac 60

ggccacaagt tcagcgtgtc cggcgagggc gagggcgatg ccacctacgg caagctgacc 120

ctgaagttca tctgcaccac cggcaagctg cccgtgccct ggcccaccct cgtgaccacc 180

ctgacctacg gcggtgcagtg cttcagccgc taccctgacc acatgaagca gcacgacttc 240

ttcaagtccg ccatgcccgaggctacgtc caggagcgca ccatcttctt caaggacgac	300
ggcaactaca agaccgcgc cgaggtgaag ttcgagggcg acaccctggt gaaccgcatc	360
gagctgaagg gcatcgactt caaggaggac ggcaacatcc tggggcacia gctggagtac	420
aactacaaca gccacaacgt ctatatcatg gccgacaagc agaagaacgg catcaagggtg	480
aatttcaaga tccgccacia catcgaggac ggagcggtgc agtcgccga ccactaccag	540
cagaacaccc ccatcggcga cgccccgtg ctgctgcccg acaaccacta cctgagcacc	600
cagtcgcccc tgagcaaaga cccaacgag aagcgcgac acatggtcct gctggagttc	660
gtgaccgccg ccgggatcac tctcggtatg gacgagctgt acaagtaaag cggccgctcg	720
agtctagagg gcccgtttaa acccgctgat cagcctcgac tgtgccttct agttgccagc	780
catctgttgt ttgccctcc cccgtgcctt ccttgacct ggaagggtgc actcccactg	840
tcctttccta ataaatgag gaaattgcat cgcattgtct gagtaggtgt cattctattc	900
tgggggggtg ggtggggcag gacagcaagg gggaggattg ggaagacaat agcaggcatg	960
ctgggggatgc ggtgggctct at	